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INFLUENCING THE SOIL MICROFLORA BY REPLACEMENT OF SPRUCE STANDS WITH MOUNTAIN ASH AND BIRCH IN THE SO_2 POLLUTED REGION

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Abstract
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In 1984 on the ridge of the Ore Mountain (sea elevation 730 m) in NW-Bohemia, an investigation into microbial colonization and soil biochemical activities in the remainder of the young spruce stand and in the grassed stands of mountain ash and birch was performed. Grassing and the development of deciduous stands contributed dramatically to the improvement of microbial conditions in the soil: the concentrations of heterotrophic bacteria were by one to two orders higher and the soil respiration, ammonification and nitrification increased. Less marked were the changes in the soil enzymatic activities. Bacterial communities from the fermentation horizon of the grassed stands had a better level of decomposing and mineralizing activities than those from the spruce stand. The results suggest that the SO_2 immissions cause no long-term soil intoxication that would hamper microbial life.

The pollution of atmosphere by sulphur dioxide devastated extensive areas of spruce stands in the Ore Mountain. These stands were replaced by cultures of different woody composition and character. A considerably large area is covered by the grassed birch and mountain ash stands. The immissions caused changes of whole forest ecosystems. A question arises, what is the influence of these changes on the soil microflora which is an integral part of the ecosystems and responds sensitively to any environmental changes.

The aim of this paper is to study the microbial colonization and soil biochemical activities in the remainder of the spruce stand without any ground vegetation and in the grassed stands of mountain ash and birch. The groups of cultures of heterotrophic bacteria, isolated from the soils of different stands, were used to study the occurrence of decomposing and mineralizing activities in the bacterial communities of these soils.

Material and methods

Three areas in the vicinity of Nová Ves in the Ore Mountain (sea elevation 730 m) were chosen for these experiments. In the first one, there is a remainder of a spruce forest stand without any groud vegetation, in the second a 15 to 20 year old thin mountain ash stand and in the third a birch stand approx. 25 years old. Both the mountain ash and the birch stands are heavily grassed (prevailingly *Calamagrostis villosa* (Chaix) J. F. Gmel). All these experimental areas are located close to each other.

Fermentation (F-A_{O2}) horizon with thickness of about 3 cm in the rest of young spruce stand is formed by decomposable material. Humus (H-A_{O3}) horizon achieves thickness of 5 to 6 cm. It is penetrated by spruce roots and contains often half decomposed remnants of ground vegetation from the period before the colsure of stand. It has a character of raw humus (mör).

In the stand of mountain ash fermentation horizon is formed by humified rests of ground vegetation and leaves of mountain ash. It is pervaded by sod roots. Its thickness is 3 cm. The share of humified material and ash moutain roots increases with depth. Humus horizon is about 7 cm thick. → H ?

In birch stand fermentation horizon is of similar character as in mountain ash. Its thickness is 6 cm. Humus horizon pervaded by birch and grass roots has a character of raw humus and is 4—6 cm thick (Lochman, unpublished).

In the course of 1984, the samples of the fermentation (F-A_{O2}), humus (H-A_{O3}) and mineral A horizons were taken separately. In the soil sieved through 2-mm screen was determined: the content of water (mass %), active pH(H_2O) and exchangeable pH(KCl) (Klika et al., 1954), content of organic carbon, total nitrogen content (mass %), ammonium nitrogen and nitrate nitrogen ($mg.kg^{-1}$) (Schlichting, Blume, 1966).

The concentrations of aerobic and ammonifying bacteria and micromycetes, determined by plating (Langkramer, Lettl, 1982), are expressed as the colony forming units (CFU) per 1 g of dry soil. The concentrations of thiosulphate oxidizers were estimated by the method of most probable number (MPN) by using liquid mineral medium for thiobacilli (Vishniac, Santer, 1957) modified in such a way that thiosulphate was the sole source of sulphur. Microbial biomass (B) was estimated according to the formula substantiated with critical qualifications by Langkramer (1977):

$$B = M.V.v.s,$$

where M — number of microorganisms in 1 g of dry soil, V — average volume of bacteria (1 μ^3) or micromycete filament (100 μ^3), v — average mass of microorganisms (= 1.1), s — dry matter of microbial mass (= 0.2). The total microbial biomass is represented by the sum of micromycete biomass and that of aerobic and ammonifying bacteria.

The soil respiration is expressed in mg of CO_2 released in 24 hrs; ammonification and nitrification in mg of NH_4^+–N or NO_3^-–N released in 14 days (Schlichting, Blume, 1966); sulphur oxidation is expressed in mg of SO_4^{2-} produced from the added elemental sulphur (0.2 $mol.kg^{-1}$) in 21 days per 1 kg of soil at 25 °C.

The concentrations of soil sacch~~o~~erase, urease (Bergmeyer, 1962), alpha-amylase (Domsch et al., 1979), asparaginase (Drobník, 1956) and thiosulphate sulphurtransferase (rhodanese; Tabatabai, Singh, 1976) are given in $kat.kg^{-1}$ ($mol.kg^{-1}.s^{-1}$).

A total number of 202 cultures of heterotrophic bacteria was isolated randomly on the meat-peptone agar from the samples of the fermentation and humus horizons of the forest soil. The cultures were tested for some of decomposing and mineralizing activities by the methods described by Lettl (1985a). Their nitrifying activity was tested on the liquid mineral medium (Pochon, 1954). The concentration of nitrate produced in 14 days was evaluated by the reaction with sodium salicylate.

Results

There are no marked differences in physical and chemical characteristics of the soils from the three above-mentioned stands. Only the soil from the spruce stand is characterized by a distinctly lower moisture. The soils from the grassed stands have a higher pH value and sorption complex saturation than the spruce stand soil (Table 1). The content of carbon, nitrogen and sulphate is practically equal in all soils (Table 2). There is, however, a significant difference in their microbial colonization. The numbers of heterotrophic bacteria are by one to two orders higher in grassed stands than those in the spruce stand. The numbers of thiosulphate oxidizers, evaluated by a different method, are similar. The numbers of micromycetes in grassed stands are also higher at the beginning and at the end of the vegetation period (Table 3). A higher colonization is confirmed also by the estimate of microbial biomass (Table 4). In the grassed stands, a higher soil respiration (Table 5), ammonification (especially in the birch stand) and nitrification (in the mountain ash stand) was observed. The differences in the content of ammonium ion are not apparent, the nitrate ion occurs in lower level in the grassed stands than in the spruce stand (Table 6). Insignificant, if any, are the differences in the ability of soil to oxidize elemental sulphur to sulphate (Table 7). As to the enzymatic activities tested, only the level of urease and sulphurtransferase (rhodanese) is higher (Table 8). Bacterial cultures, isolated from three types of stands, were tested tentatively for some decomposing and mineralizing activities. It appears, that the bacterial communities of grassed stands are better equipped with these activities than the communities of the spruce stand. This is valid only for the fermentation horizons, not for the humus ones (Table 9). Most of the heterotrophic bacteria had the nitrifying activity; the highest effectivity was found in the isolates from the spruce stand soil (Table 10).

Discussion

This paper includes the preliminary results of the comparison of soil microflora from three types of stands, performed in 1984. Therefore these results can serve only for orientation. Nevertheless, they suggest that grassing and planting of deciduous stands, following the spruce stand, had a dramatic positive impact on the microbial conditions in the soil: the soil colonization is much higher, bacterial communities from the fermentation horizons are much better equipped with decomposing and mineralizing activities than those from the spruce stand soil. Also the soil activities from the carbon and nitrogen cycle are markedly higher. The above-mentioned changes can have different reasons.

In comparison with the spruce stand, the soil moisture increased in deciduous stands. This is probably due to a high interception of spruce stand which keeps a considerable proportion of precipitations in the canopy of crowns (Table 1).

There was an increase in the extremely acid soil pH value and in the saturation of the sorption complex. The grasses and the deciduous stand take up larger amount of cations from deeper soil layers which then reach the upper soil horizons with the dead plant matter (Lettl, Lochman, 1984). Moreover, ammonification contributes to the changes in the soil pH value by the formation of ammonium ion. Therefore the average pH value in the fermentation horizons of the deciduous stands was by 0.8, in

Table 1. Moisture (mass percent) and pH of soils from three types of forest stands

Horizon	Sampling in 1984	Moisture [%]			pH(H_2O)			pH(KCl)		
		spruce	mountain ash	birch	spruce	mountain ash	birch	spruce	mountain ash	birch
F-A$_{02}$	May 10	54.6	64.2	63.4	3.3	5.6	4.0	3.1	4.9	3.6
	June 21	55.6	64.0	67.6	3.6	3.9	4.0	3.3	3.5	3.6
	July 10	57.1	63.3	56.4	3.3	4.3	4.2	2.6	3.5	3.5
	September 10	63.4	68.6	71.9	3.6	3.6	4.7	3.1	3.1	4.0
	October 30	56.1	64.1	64.7	3.5	4.0	4.2	3.2	3.5	3.6
	December 5	64.5	68.3	69.8	3.6	4.4	4.5	3.3	3.7	3.8
H-A$_{03}$	May 10	56.3	62.8	61.8	3.4	4.0	3.9	2.9	3.3	3.5
	June 21	54.9	59.2	62.4	3.4	3.8	3.6	2.8	3.1	3.1
	July 10	54.9	61.7	43.9	3.2	3.9	3.7	2.9	3.4	3.1
	September 10	57.9	64.4	58.9	3.4	4.0	3.9	3.0	3.4	3.4
	October 30	52.8	59.5	58.2	3.4	3.7	3.7	2.9	3.3	3.4
	December 5	54.5	66.2	60.7	3.7	4.2	3.9	3.2	3.6	3.4
A	May 10	31.7	24.8	34.7	3.6	3.7	3.6	3.2	3.0	3.3
	June 21	30.0	32.2	35.2	3.2	3.8	3.8	2.7	2.9	3.2
	July 10	25.3	31.8	33.4	3.5	3.5	3.7	2.7	2.9	3.1
	September 10	33.3	27.5	25.2	3.6	3.6	3.7	2.8	3.3	3.3
	October 30	33.4	33.8	33.1	3.9	4.0	3.7	3.6	3.0	3.1
	December 5	29.5	25.8	35.4	3.2	3.6	3.8	2.9	3.3	3.3
F-A$_{02}$	$\bar{x}$	58.5	65.4	65.3	3.48	4.30	4.27	3.10	3.70	3.68
	s_x	4.27	2.37	5.52	0.15	0.70	0.28	0.26	0.62	0.18
H-A$_{03}$	$\bar{x}$	55.2	62.5	57.6	3.42	3.93	3.78	2.95	3.35	3.32
	s_x	1.73	2.75	6.93	0.16	0.17	0.13	0.14	0.16	0.17
A	$\bar{x}$	30.5	29.3	32.8	3.50	3.70	3.72	2.98	3.07	3.22
	s_x	3.03	3.76	3.86	0.27	0.18	0.07	0.35	0.19	0.10

Table 2. Content of organic carbon, of total nitrogen (mass percent) and sulfate content ($\mu g \cdot g^{-1}$ over dry soil) in soils of three types of forest stands

Horizon	Sampling in 1984	C_{ox} [%]			N_{total} [%]			C/N			SO_4^{2-} [$\mu g \cdot g^{-1}$]		
		spruce	mountain ash	birch	spruce	mountain ash	birch	spruce	mountain ash	birch	spruce	mountain ash	birch
F-A$_{02}$	May 10	33.9	31.7	33.0	1.56	1.65	1.74	21.7	19.2	18.9	294	259	208
	June 21	33.4	30.0	34.4	1.80	1.43	1.62	18.6	21.0	21.3	264	211	311
	July 10	32.4	32.9	34.1	1.42	1.46	1.84	22.8	22.5	18.5	259	246	222
	September 10	30.4	34.7	28.3	1.74	1.62	1.44	17.5	21.4	19.6	388	288	329
	October 30	28.8	30.7	31.7	1.38	1.36	1.48	20.8	22.6	21.4	225	275	554
	December 5	33.2	33.1	32.3	1.58	1.68	1.60	21.0	19.7	20.2	585	364	518
H-A$_{03}$	May 10	25.5	30.5	33.0	1.10	1.20	1.36	23.3	25.4	24.1	84.7	166	145
	June 21	21.5	30.3	30.8	1.00	1.08	1.34	21.5	27.9	23.0	214	141	120
	July 10	23.4	30.1	24.6	1.06	1.30	1.05	22.0	23.2	23.4	269	161	176
	September 10	29.8	24.3	21.5	1.22	1.29	1.30	24.4	18.9	16.5	352	191	145
	October 30	25.5	23.8	24.0	0.92	1.08	1.62	27.8	22.1	14.8	205	183	222
	Debember 5	24.0	30.3	27.2	1.08	1.12	1.16	22.2	27.1	23.4	489	305	393
A	May 10	7.49	4.43	6.92	0.35	0.16	0.28	21.4	27.0	24.4	—	—	—
	June 21	4.70	5.22	6.51	0.19	0.18	0.28	25.0	28.5	23.1	—	—	—
	July 10	3.76	4.72	7.75	0.17	0.18	0.45	22.6	26.2	17.2	—	—	—
	September 10	7.11	5.13	5.83	0.29	0.22	0.22	24.7	23.1	25.9	—	—	—
	October 30	4.51	5.65	6.01	0.17	0.19	0.27	27.0	30.2	22.2	—	—	—
	Debember 5	4.45	4.49	7.89	0.17	0.13	0.29	26.2	33.5	26.8	—	—	—
F-A$_{02}$	$\bar{x}$	32.0	32.2	32.3	1.58	1.53	1.62	20.4	21.1	20.0	336	274	357
	s_x	2.00	1.73	2.21	0.17	0.13	0.15	1.98	1.41	1.21	134	51.5	147
H-A$_{03}$	$\bar{x}$	24.9	28.2	26.8	1.06	1.18	1.30	23.5	24.1	20.9	269	191	200
	s_x	2.80	3.23	4.37	0.10	0.10	0.19	2.33	3.38	4.09	139	58.4	101
A	$\bar{x}$	5.34	4.94	6.82	0.22	0.18	0.30	24.5	28.1	23.3	—	—	—
	s_x	1.56	0.47	0.87	0.08	0.03	0.08	2.13	3.56	3.43	—	—	—

Table 3. Numbers of microorganisms (in thousands CFU or MPN per g soil) in soils of three forest stands (CFU — colony forming units; MPN — most probable number)

Horizon	Sampling in 1984	Aerobic bacteria (CFU)			Ammonifying bacteria (CFU)			$S_2O_3^{2-}$ oxidizers (MPN)			Microfungi (CFU)		
		spruce	mountain ash	birch	spruce	mountain ash	birch	spruce	mountain ash	birch	spruce	mountain ash	birch
F-A$_{02}$	**May** 10	169	11359	2668	382	17616	3707	24.2	2570	355	176	689	300
	June 21	954	2812	6422	1209	4809	7636	383	777	6792	188	157	720
	July 10	589	39769	12786	1196	36374	26153	6513	435727	367057	186	154	291
	September 10	1970	5159	377790	4624	9108	471291	9302	5095	569157	830	297	237
	October 30	1016	4026	92645	1016	4128	97258	6372	7236	48213	728	408	151
	December 5	544	16582	10022	1220	27441	17218	6758	11048	11589	103	484	717
H-A$_{03}$	**May** 10	554	1120	1414	1381	1918	2583	0.46	296	62.8	99.2	260	244
	June 21	1034	710	1727	1264	972	2480	10.8	318	3454	14.8	65.3	239
	July 10	487	1956	945	960	2295	1088	2881	57381	23189	7.39	52.2	113
	September 10	783	1965	2764	1614	2934	4474	33.2	449	2067	111	103	130
	October 30	1171	1383	2818	1369	815	4168	1439	543	4096	212	255	327
	December 5	601	1431	3070	1115	3139	3299	37.4	977	356	44.0	474	246
A	**May** 10	58.6	31.0	245	107	120	434	—	—	—	24.4	4.63	15.3
	June 21	109	152	103	152	98.4	56.6	—	—	—	4.76	0	5.14
	July 10	53.5	48.9	295	66.9	127	240	—	—	—	4.46	0	10.0
	September 10	85.0	138	71.3	135	396	31.2	—	—	—	5.00	13.8	17.8
	October 30	490	90.6	124	515	201	154	—	—	—	10.0	35.2	9.96
	December 5	113	62.9	46.4	137	153	217	—	—	—	18.9	31.4	25.8
F-A$_{02}$	$\bar{x}$	875	13284	83722	1608	16579	110544	4892	77075	167194	368	365	403
	s_x	618	13982	148030	1502	13117	183385	3789	175739	242261	321	206	250
H-A$_{03}$	$\bar{x}$	772	1427	2123	1284	2014	3015	735	9994	5537	81.4	202	216
	s_x	278	486	876	228	972	1243	1195	23216	8796	76.9	162	80.7
A	$\bar{x}$	151	87.2	147	185	183	189	—	—	—	11.2	14.2	14.0
	s_x	168	49.0	99.8	164	110	146	—	—	—	9.43	15.7	7.30

Table 4. Total biomass of microorganisms in soils

Horizon	Sampling in 1984	Total microbial biomass [mg]								
		per kg soil			per 10 g C_{ox}			per 10 g N_{total}		
		spruce	mountain ash	birch	spruce	mountain ash	birch	spruce	mountain ash	birch
F-A$_{02}$	May 10	4.00	22.1	8.08	0.12	0.70	0.24	2.56	13.4	4.64
	June 21	4.70	5.30	19.8	0.14	0.18	0.58	2.61	3.71	12.2
	July 10	5.92	116	95.8	0.18	3.53	2.81	4.17	79.4	52.1
	September 10	21.8	10.8	317	0.72	0.31	11.2	12.5	6.67	220
	October 30	18.1	12.4	55.7	0.63	0.40	1.76	13.1	9.12	37.6
	December 5	4.14	22.8	24.3	0.12	0.69	0.75	2.62	13.6	15.2
H-A$_{03}$	May 10	2.61	6.45	6.26	0.10	0.21	0.19	2.37	5.37	4.60
	June 21	0.83	1.88	6.94	0.04	0.06	0.22	0.83	1.74	5.18
	July 10	1.11	14.7	8.03	0.05	0.45	0.33	1.05	0.67	7.65
	September 10	2.98	3.44	4.91	0.10	0.14	0.23	2.44	2.67	3.78
	October 30	5.54	6.21	9.63	0.22	0.26	0.40	6.02	5.75	5.94
	December 5	1.35	12.0	6.89	0.06	0.40	0.25	1.25	10.7	5.94
A	May 10	0.57	0.13	0.49	0.08	0.03	0.07	1.62	0.81	1.75
	June 21	0.16	0.06	0.15	0.03	0.01	0.02	0.84	0.33	0.53
	July 10	0.12	0.04	0.34	0.03	0.01	0.04	0.71	0.22	0.75
	September 10	0.16	0.42	0.41	0.02	0.08	0.07	0.55	1.91	1.86
	October 30	0.44	0.84	0.28	0.10	0.15	0.05	2.59	4.42	1.04
	December 5	0.47	0.74	0.62	0.11	0.16	0.08	2.76	5.69	2.14
F-A$_{02}$	$\bar{x}$	9.78	31.6	86.8	0.32	0.97	2.89	6.26	21.0	57.0
	s_x	7.99	41.9	117	0.28	1.27	4.18	5.11	28.9	81.8
H-A$_{03}$	$\bar{x}$	2.40	7.45	7.11	0.09	0.85	0.27	2.33	4.48	5.51
	s_x	1.74	4.96	1.60	0.07	0.15	0.08	1.93	3.64	1.33
A	$\bar{x}$	0.32	0.37	0.38	0.06	0.07	0.05	1.51	2.23	1.34
	s_x	0.19	0.35	0.16	0.04	0.07	0.02	0.97	2.30	0.66

Table 5. Basal (B) and potential (G) respiration (in mg CO_2 per kg soil in 24 h) of soils from three types of forest stands

Horizon	Sampling in 1984	Basal respiration (B)			Potential respiration (G)			G/B		
		spruce	mountain ash	birch	spruce	mountain ash	birch	spruce	mountain ash	birch
F-A_{02}	May 10	>3793	>4810	4316	6082	>9621	9411	—	—	2.18
	June 21	3792	>5372	5127	7501	>10744	11487	1.98	—	2.24
	July 10	1075	733	2727	3663	4098	7910	3.41	5.59	2.90
	September 10	2155	2817	>6386	6366	6838	>12773	2.95	2.43	—
	October 30	2381	3106	4360	3621	8993	>10372	1.52	2.90	—
	December 5	2222	3700	4339	6309	4919	10542	2.84	1.33	2.43
H-A_{03}	May 10	1003	2455	1319	2928	6420	2179	2.92	2.61	1.65
	June 21	710	1046	1793	1910	2458	4339	2.69	2.35	2.42
	July 10	887	1999	1066	1891	3851	4216	2.13	1.93	3.96
	September 10	829	1630	1288	3802	3459	3509	4.58	2.12	2.72
	October 30	1263	1659	1914	6237	2870	4544	4.94	1.73	2.37
	December 5	1203	1962	1921	3721	3603	4940	3.09	1.84	2.57
A	May 10	146	104	207	322	365	503	2.25	3.51	2.42
	June 21	191	328	489	510	597	682	2.67	1.82	1.39
	July 10	262	437	605	>2586	2187	2692	—	5.01	4.44
	September 10	126	180	201	356	457	390	2.83	2.53	1.94
	October 30	532	444	467	687	1552	991	1.29	3.50	2.12
	December 5	347	349	485	860	715	1012	2.48	2.05	2.08
F-A_{02}	$\bar{x}$	—	—	—	5590	—	—	—	—	—
	s_x	—	—	—	1588	—	—	—	—	—
H-A_{03}	$\bar{x}$	982	1792	1550	3415	3777	3954	3.39	2.10	2.61
	s_x	217	471	370	1614	1849	988	1.11	0.33	0.75
A	$\bar{x}$	267	307	409	—	979	1045	—	3.07	3.40
	s_x	153	138	166	—	728	845	—	1.19	1.52

Table 6. Soil activities from the nitrogen cycle (in mg NH_4^+–N and NO_3^-–N per kg soil in 14 d) and content of their products (in mg NH_4^+–N and NO_3^-–N per kg soil)

Horizon	Sampling in 1984		Ammonification			NH_4^+–N content			Nitrification			NO_3^--N content		
			spruce	mountain ash	birch	spruce	mountain ash	birch	spruce	mountain ash	birch	spruce	mountain ash	birch
F-A_{02}	May	10	202	426	465	113	156	116	0	93.8	0	0	0	0
	June	21	131	218	346	85.8	124	138	0	0	0	0	0	0
	July	10	73.1	97.7	175	131	232	159	10.2	21.8	16.5	4.66	0	1.84
	September	10	153	178	438	196	64.2	191	4.38	30.6	0	0	0	0
	October	30	138	193	299	76.5	99.8	127	45.5	2.23	6.81	29.1	8.91	9.07
	December	5	0	84.9	89.1	366	156	453	0	10.1	0	18.0	10.1	5.30
H-A_{03}	May	10	108	349	129	118	48.2	46.9	0	0	20.9	0	0	0
	June	21	69.5	71.4	155	49.6	38.4	59.5	0	0	0	0	0	0
	July	10	0	23.4	48.0	114	93.5	68.0	6.20	12.5	11.4	7.98	0	0
	September	10	58.5	88.1	131	26.6	44.0	16.3	0	40.4	17.5	0	0	0
	October	30	33.2	0	107	38.0	77.5	75.2	8.47	15.8	0	22.0	7.90	13.4
	December	5	0	0	5.71	251	246	222	0	2.37	0	10.6	2.37	4.07
A	May	10	32.8	25.3	22.3	6.56	4.47	6.86	0	0	0	0	0	0
	June	21	11.2	18.2	12.1	14.4	11.6	12.1	0	0	0	0	0	0
	July	10	0	0	6.74	15.0	23.0	10.1	4.28	4.69	13.8	0	0	0
	September	10	10.1	6.19	5.99	1.68	4.64	2.99	0	0	0	0	0	0
	October	30	0	0	1.67	32.0	11.8	15.1	2.40	0	0	4.20	1.81	5.38
	December	5	0	0	0	55.6	46.8	53.7	0	0	0	3.97	2.70	0
F-A_{02}	$\bar{x}$		116	200	302	161	139	197	10.0	26.4	6.63	8.63	3.17	2.70
	s_x		82.7	123	147	109	57.6	128	17.8	35.0	7.39	12.2	4.92	3.74
H-A_{03}	$\bar{x}$		44.9	88.6	95.9	99.5	91.3	81.3	2.44	11.8	8.30	6.76	1.71	2.91
	s_x		42.3	133	57.2	83.8	78.8	72.0	3.85	15.5	9.59	8.78	3.18	5.39
A	$\bar{x}$		9.02	7.78	8.13	20.9	17.0	16.8	1.11	0.78	2.30	1.36	0.75	0.90
	s_x		12.8	10.9	8.13	19.9	16.1	18.5	1.82	1.91	5.63	2.11	1.20	2.20

Table 7. S^0 oxidation (in mg SO_4^{2-} per kg soil in 21 d) in soils from three types of forest stands

Horizon	Sampling in 1984		Oxidation of S^0 to SO_4^{2-}		
			spruce	mountain ash	birch
F-A_{02}	May	10	974	7280	3176
	June	21	5510	4515	5881
	July	10	7617	4985	4242
	September	10	7964	5614	2554
	October	30	5543	4843	3523
	December	5	7369	5370	5129
H-A_{03}	May	10	1365	2527	3360
	June	21	4598	2671	5470
	July	10	2201	3884	3177
	September	10	3619	3303	2686
	October	30	3279	2632	2833
	December	5	4558	4605	2664
F-A_{02}	$\bar{x}$		5829	5434	4084
	s_x		2603	984	1251
H-A_{03}	$\bar{x}$		3270	3270	3365
	s_x		1290	834	1068

the humus horizons by 0.4–0.5 and in the mineral A horizons by 0.2 higher than that in the identical horizons of the spruce stand (Table 1).

As observed further, the grassed deciduous stands offer more valuable nutritive substrate to the soil microflora in litter. There are no apparent changes in the content of carbon and nitrogen (Table 2), however, there are probably significant differences in the qualitative composition of organic matter. The shedded leaves of deciduous trees and the mass of dead grasses are obviously more susceptible to microbial attack than the shedded needles (more favourable composition of organic matter, higher content of saccharides, absence of resins and essential oils with antibiotic effect, etc.). The decisive factor seems to be the grass rhizosphere. Root exudates contain very valuable and easily metabolizable nutritive substances (saccharides, amino acids, etc.); their quantity can be surprisingly high (Přikryl, Vančura, 1980). The high nutritional value of organic matter in the grassed deciduous stands is supported by the fact that these soils have higher bacterial counts, and mainly by the fact that the unit amount of organic matter (expressed as C_{ox} or N_{total}) contains a larger amount of microbial biomass (Table 4), although its mathematical estimate is dubious.

Easier decomposability of the grass stand material is documented by the value of soil respiration, which is considered as a total indicator of organic matter decomposition. The respiration in the fermentation horizon often surpassed the possibilities of

Table 8. Concentration of enzymes in forest soils

Horizon	Sampling in 1984	Saccharose [mkat . kg^{-1}]			Alpha-amylase [mkat.kg^{-1}]			Urease [nkat . kg^{-1}]			Asparaginase [nkat . kg^{-1}]			Rhodanese [nkat . kg^{-1}]		
		spruce	mountain ash	birch	spruce	mountain ash	birch	spruce	mountain ash	birch	spruce	mountain ash	birch	spruce	mountain ash	birch
F-A$_{02}$	May 10	2.98	0.99	1.20	0.86	0.69	0.60	0.64	2.31	0.86	0.11	0.67	0.11	0.81	14.2	23.1
	June 21	1.34	1.10	1.25	0.90	0.47	0.91	0.72	0.97	1.17	0.94	0.39	0.05	0.61	6.11	1.94
	July 10	1.29	1.28	1.16	0.42	0.33	0.35	0.44	1.97	0.83	0.25	0.75	0.89	2.63	3.93	2.13
	September 10	2.51	2.15	1.78	0.66	0.53	0.65	0.36	0.08	0.17	0.25	0.17	0.61	2.02	14.0	1.21
	October 30	1.24	0.60	2.11	0.50	0.15	0.69	0.67	1.47	1.11	0.58	0.11	0.55	0	0	5.67
	December 5	1.21	2.54	1.80	0.65	0.26	0.51	0.25	1.28	0.31	0.50	0.42	1.11	3.65	8.10	4.45
H-A$_{03}$	May 10	1.63	1.17	1.04	0.39	0.51	0.56	0.85	0.86	0.36	0.05	0.42	0.17	4.05	4.05	4.86
	June 21	1.18	1.33	1.19	0.42	0.58	0.73	0.30	0.75	1.25	0.67	0.08	0.25	5.10	3.42	0.53
	July 10	1.31	1.25	1.27	0.36	0.31	0.31	0.25	1.55	0.94	0.11	0.44	0.14	3.23	7.41	0.61
	September 10	1.30	1.67	1.89	0.33	0.46	0.56	0.03	0.08	0.11	0.001	0.58	0.45	4.25	7.70	0.81
	October 30	0.78	1.17	1.42	0.34	0.33	0.62	0.11	0.22	0.75	0.39	0.42	0.28	1.42	2.02	1.62
	December 5	1.50	1.71	1.44	0.48	0.49	0.64	0.17	0.22	0.83	0.58	0.64	0.53	3.65	4.05	1.62
F-A$_{02}$	$\bar{x}$	1.76	1.44	1.55	0.66	0.40	0.62	0.51	1.35	0.74	0.44	0.42	0.55	1.62	7.72	6.42
	s_x	0.77	0.74	0.40	0.19	0.20	0.19	0.19	0.78	0.41	0.30	0.26	0.42	1.39	5.62	8.35
H-A$_{03}$	$\bar{x}$	1.28	1.38	1.37	0.39	0.45	0.57	0.28	0.61	0.71	0.30	0.43	0.30	3.62	4.77	1.67
	s_x	0.29	0.24	0.29	0.06	0.11	0.14	0.29	0.57	0.44	0.29	0.19	0.15	1.25	2.30	1.63

Table 9. Occurrence of biochemical activities in heterotrophic bacteria isolated on meat-peptone agar from forest soils

Type of forest stand	Spruce			Mountain ash		Birch
Date of sampling	Oct 5, 1982	Jan 12, 1983	May 10, 1984	Oct 5, 1982	May 10, 1984	May 10, 1984
Horizon F-A_{02}						
Number of isolates	24	25	21	28	22	23
Percent of isolates with the activity						
amylolysis	37.5	36.0	66.7	96.4	86.4	52.2
proteolysis	8.3	0	57.1	96.4	72.7	69.6
ammonification of asparagine	62.5	96.0	95.2	100.0	95.4	100.0
oxidation of NH_4^+ to NO_3^-	0	64.0	95.2	64.3	100.0	90.3
oxidation of NO_2^- to NO_3^-	58.3	—	33.3	71.4	90.9	90.3
reduction of S^0 to S^{2-}	29.2	4.0	28.6	92.9	31.8	65.2
oxidation of S^0 to SO_4^{2-}	91.7	88.0	85.7	100.0	63.6	69.2
oxidation of $S_2O_3^{2-}$ to SO_4^{2-}	25.0	0	—	100.0	—	—
production of sulfurtransferase	8.3	—	—	92.9	—	—
production of thiosulfate oxidase	29.2	—	—	64.3	—	—
production of sulfite oxidase	8.3	—	—	25.0	—	—
Horizon H-A_{03}						
Number of isolates	—	—	22	—	13	24
Percent of isolates with the activity						
amylolysis	—	—	95.4	—	69.2	91.7
proteolysis	—	—	54.5	—	53.8	70.8
ammonification of asparagine	—	—	80.9	—	84.6	75.0
oxidation of NH_4^+ to NO_3^-	—	—	100.0	—	100.0	91.7
oxidation of NO_2^- to NO_3^-	—	—	90.9	—	100.0	100.0
reduction of S^0 to S^{2-}	—	—	18.2	—	7.7	37.5
oxidation of S^0 to SO_2^{2-}	—	—	81.8	—	92.3	83.3

Table 10. Rate of oxidation of NH_4^+ (in nmol/L NO_3^-) and that of NO_2^- (in mmol/L NO_3^- produced in 14 d) in heterotrophic bacteria isolated from forest soils (sampling on May 10, 1984)

Type of forest stand		Spruce		Mountain ash		Birch	
Horizon		F—A_{02}	H-A_{03}	F-A_{02}	H-A_{03}	F-A_{02}	H-A_{03}
Number of isolates		21	22	22	13	23	24
Oxidation of NH_4^+ to NO_3^- (NO_3^-; nmol/L/14 d)	Number of nitrifiers	20	22	22	13	21	22
	Rate of oxidation $\bar{x}$	20.2	19.1	12.4	9.15	10.0	14.4
	s_x	6.50	6.48	7.41	4.37	6.05	7.23
Oxidation of NO_2^- to NO_3^- (NO_3^-; nmol/L/14 d)	Number of nitrifiers	7	20	20	13	21	24
	Rate of oxidation $\bar{x}$	1.93	4.59	1.13	1.54	1.38	5.72
	s_x	2.22	1.99	0.46	0.65	1.01	1.29

the employed method. In the course of the year, the respiration in deciduous stands was always higher than in the spruce stand. In different soils, addition of 1 % glucose had a different effect. As a rule, in the spruce stand soil the CO_2 production increased dramatically (high ratio G:B), in the grass stands this effect was less apparent (Table 5). This suggests that the soil from the grassed stands contained a higher level of the low-molecular and easily utilizable saccharides.

As another indicator of the organic matter decomposability may serve the mineralization of nitrogen. Ammonification is stimulated extensively in the grassed stands, the highest mean values being observed in the birch stand. This activity is obviously suppressed by the produced ammonium ion because it starts as soon as the ammonium ion level in the soil decreases (Table 6).

Ammonium ion serves as an oxidizable substrate for nitrification. This activity was proved mostly during the summer months and the best conditions for it were in the grassed mountain ash stand. Nitrates appeared in the soil only towards the end of the vegetation period, their level being substantially lower in the grassed stands than in the spruce stand (Table 6). Their uptake from the soil occurred most probably through the root system of grass vegetation than by rainfall wash-out.

As to the elemental sulphur oxidation, there were only small differences in the fermentation horizons which were disappearing already in the humus horizons (Table 7). Similar small, and not easily explainable differences were observed in the soil enzymatic activities. In the grassed stands, higher levels were determined only with urease and sulphurtransferase (rhodanese) (Table 8).

The type of vegetation, i.e. that of the organic matter, also influenced the occurrence of decomposing and mineralizing activities in the communities of soil heterotrophic bacteria. The composition of these communities is extremely variable according to the seasonal type of available substrate. In bacterial communities from the fermentation horizon of the grassed stands there was a higher occurrence of active cultures than in the communities from the spruce stand (Table 9). These differences can be seen quite clearly in the samples taken in October, much less in those taken in May. There were no distinct differences in the isolates from the humus horizons. It is therefore probable that the organic material in the fermentation horizon of the grassed deciduous stands is more diverse, which results in structural changes of bacterial communities and in induction of numerous activities. Microflora in these stands is much better equipped with biochemical activities necessary for maintaining healthy soil.

These analyses proved two microbiological specialities of the studied locality. The first one is a high occurrence of the nitrifying activity in bacteria. Heterotrophic nitrification in bacteria is well known (Eylar, Schmidt, 1959), however, it was estimated as negligible. In acid soils only the heterotrophic nitrification can take place. Only a small amount of nitrate was produced in conditions of the test in vitro (Pochon, 1954). Moreover, the nitrifying bacteria from the grassed stands were less effective than the cultures from the spruce stand (Table 10), though the situation was quite opposite with the soil nitrification (compare with Table 6). This suggests that at excessive multiplication of bacterial microflora by one to two orders, a substantially lower amount of substrate (ammonium ion) is available per unit amount of microorganisms, which means that the production of corresponding enzymatic systems is stimulated only in a limited way.

The second speciality is a high occurrence of heterotrophic oxidizers of sulphur. Thiobacilli are considered as the main oxidizers, but in the regions rich in sulphur such activity is induced also in heterotropic organisms. This phenomenon was observed in the regions polluted by soot (Wainwright, 1978b), by spreading of sulphur in the vicinity of sulphur mines (Król, 1983), or by SO_2 immissions (Lettl, 1984).

Let us consider the importance of these findings for judging the conditions of forest soil.

The study on the influence of SO_2 immisions on the soil microflora of coniferous (prevailingly spruce) stands revealed a number of negative, though not too apparent, impacts (Mrkva, Grunda, 1969; Langkramer, Lettl, 1982). These facts, and mainly mass dying off of spruce, led to the conclusion that there may exist a long-term intoxication, causing at least local disappearance of microbial life and biochemical activities in the soil (Muranský, 1983). Let us consider if such intoxication is possible.

In spite of the fact that some areas are more exposed to immissions of some heavy metals, the so far determined concetrations could not have had any negative impact on microflora (Lochman, Lettl, 1984).

Acidification, i. e. the increase in H^+ ion concentration in organic horizons of the polluted soils was not proved in our plots (Langkramer, Lettl, 1982), and therefore it was impossible to substantiate the negative impact of immissions on microflora by this mechanism (Lettl, 1985a). In this respect, the spruce stand itself and the sea elevation of the locality play a more important role. It is known that microflora has adapted itself to the acid environment by developing acidophilic communities which seem practically equal to neutrophilic ones (Lettl, 1984).

The possibility of intoxication by sulphur should be considered. In the soil of polluted areas high levels of sulphur cycle intermediates (Wainwright, 1978a), a high content of sulphur in the form of solid deposites (Wainwright, Killham, 1982) and/or in the bound to humic acids (Kerndorff, Schnitzer, 1979) were proved. The oxidation of this sulphur supply proceeds only slowly (Killham, Wainwright, 1984). This sulphur represents therefore either a very stable or a very substantial soil component. The following adaptations of microbial communities were observed: a higher proportion of resistant cultures, higher concentration of thiobacilli, induction of ability of sulphur oxidation also in heterotrophic microflora (Wainwright, 1978b; Król, 1983; Lettl, 1984). This indirectly excludes the possibility of explicit toxic behaviour of any intermediates in the aerated soil environment. It is also impossible to take into consideration the toxicity caused by SO_3^{2-}/HSO_3^-. Its persisting concentrations in the soil are by far not toxic (Wainvright, Johnston, 1980). As known, low concentrations can have rather a stimulating effect (Lettl, 1984). Microflora in the areas exposed to immissions is damaged only in the uppermost fermentation horizon but not in the humus horizon. Toxic influence of immissions can be therefore attributed mostly to the direct influence of gaseous SO_2 (Lettl, 1985b).

The soil in the polluted areas can be considered rather to be under a long-term influence of excessive sulphur supply than under long-term intoxication. The intoxicated soil would not permit such a strong development of microflora as can be seen under improved nutritional conditions in the grassed deciduous stands. In the areas exposed permanently to SO_2 the occurrence of different microbial communities (resistant, acidophilic, other species structure, etc.) than in the unpolluted areas was observed. The communities from grassy deciduous stands are characterized by a higher number of activities necessary for maintaining soil functions. Practically, soil intoxication does not develop even in the surroundings of the sulphur mines in Poland where sulphur level varied from 0.8 up to 3.9 %. Not even the lowest sulphur concentration admitted any existence of vegetation and microbial colonization was negligible. The pH value in the soil amounted up to 1.4, however, mere mass liming renewed not only the microbial activity, but also the soil fertility, as proved by pot trials with wheat (Król et al., 1972). As apparent, the negative factor is not the sulphur itself, but the low pH value resulting from the oxidation of sulphur to sulphate in insufficiently buffered environment. Liming is also not without problems because it can bring about a temporary nitrogen immobilization. Moreover, even if liming has a positive effect on the pH value and soil microflora, the forest growth is influenced rather by intensive mechanical treatment of soil than by liming itself (Palmgren, 1984).

We are well aware of presenting a simplified explanation of these problems, however, no better alternative is available at present. The recent state of information and the current results suggest that lowering the SO_2 concentration in the atmosphere below the toxicity limit would enable the utilization of this soil as forest soil, provided the loss of cations could be balanced. As follows from this study, no symptoms of permanent or long-term soil intoxication were proved.

Translation submitted by the author

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Lettl A.: **Ovlivnění půdní mikroflory náhradou smrkových porostů jeřábem a břízou v oblasti znečištěné imisemi SO_2.**

Na hřebeni Krušných hor (730 m n. m.) v severozápadních Čechách bylo v r. 1984 sledováno mikrobní osídlení a půdní biochemické aktivity ve zbytku mladého smrkového porostu a v zatravněných porostech jeřábu a břízy. Zatravněním a rozvojem listnatých porostů byly dramaticky zlepšeny mikrobiální poměry v půdě: koncentrace heterotrofních baktérií byly o jeden až dva řády vyšší, zvýšila se půdní respirace, amonizace a nitrifikace. Méně výrazné byly změny půdních enzymatických aktivit. Bakteriální společenstva z fermentačního horizontu zatravněných porostů byla lépe vybavena rozkladnými a mineralizačními aktivitami než společenstva porostu smrkového. Výsledky naznačují možnost, že imise SO_2 nezpůsobují dlouhodobou intoxikaci půdy, která by znemožnila mikrobiální život.

Леттл А.: **Влияние замены еловых насаждений рябиной и березой в области загрязненной имиссиями SO_2 на почвенную микрофлору.**

На гребени Крушных гор (730 м н. у. м.) в северозападной Чехии было в 1984 г. исследованно микробное заселение и почвенные биохимическийе активности в остатке молодого елового насаждения и в затравянелых насаждениях рябины и березы. Затравянением и развитием лиственных насаждений драматически улучшились микробиальные условия в почве: концентрации гетеротрофных бактерий были на один, два порядка больше, повысилась почвенная респирация, амонизация и нитрификация. Меньше яркими являлись изменения почвеных энзиматических активностей. Бактериальные сообщества из ферментационного горизонта затравянелых насаждений были лучше снабжены разлагающими и минерализационными активностями чем сообщества насаждения елового. Результаты намечают возможность, что имиссии SO_2 не вызывают долговременную интоксикацию почвы, которая бы сделала невозможной микробиальную жизнь.